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SUMMARY OF DOCTORAL THESIS

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Title: AGRICULTURAL BIOFORTIFICATION OF RICE THROUGH MICRONUTRIENTS AVAILABILITY CONTROL AND VARIETAL SELECTION.

(土壌の微量要素可給性の制御と品種選択による米の農業生物学的栄養強化)

This study examined the feasibility of agricultural practices (irrigation and fertilization and selection of varieties) to increase the concentration of micronutrients in rice grain, emphasizing on effect of water management and timing to control the availability of micronutrients in soil. The study further compares genotypic differences between upland and lowland rice varieties in term of nutrients and phytic acid concentration in rice grain.

In an exploratory study, the effects of water saving strategy on rice micronutrient concentration during post-heading stage in two soil types (Typic Fluvaquent and Typic Paleudult) was assessed, as well as its impact on morphology, yield parameters and availability of micronutrients in soil. The results revealed that regardless of soil type, the establishment of water saving did not have negative effects on morphological traits, meanwhile the grain yield was kept comparable to the conventional cultivation patterns. Establishment of aerobic soil condition during post anthesis stage also reduced the grain sterility, as well as increased root and shoot dry-mass in both soil. These features not only assisted rice water use efficiency, but also have potential of increasing micronutrient uptake efficiency due to its enhanced in root-soil contact. Changes in soil electrochemistry (pH and redox potential) also played an important role controlling micronutrient availability and toxicity. It was observed that availability of Zn slightly increased with the establishment of aerobic conditions while the toxic level of Mn in Typic Paleudult was reduced to less harmful levels. Mn toxicity was responsible for poor morphological behavior with consequent reduction in yield parameters and feasibility of this soil for rice production. In summary, the water saving practice in the post-anthesis stage has the potential to improve water use efficiency and to control micronutrients availability in soil, without sacrificing yield parameters.

In subsequent study of water saving strategy, the degree of effect of the availability of micronutrient in soil on the biofortification of rice grain was significantly varied at different reproductive stages (anthesis to dough-milky grain). Changes in soil redox potential and pH, from two weeks after heading, altered concentration of Zn, Cu, and Mn in grain. Although rice Fe concentration did not suffer considerable changes as consequence of water management, probably soil re-oxidation brought Fe to less available form, reducing the antagonist reactions with aforementioned elements. The grain-filling stage seems to be the most active stage for micronutrient uptake and translocation to the grain. Timing at this stage can be taken as reference for any biofortification purpose. Thus, changes in soil electrochemical properties

through implementation of water management strategies seems to be the most feasible and environmentally friendly way to increase availability of micronutrients in soil and the concentration in grain.

The application of farmyard manures (FYM) and other forms of organic matter contributed to soil micronutrients availability by changing physical, chemical, and biological soil properties. The combination of NPK-FYM was the best approach for improving biomass, number of panicles, yield and reduction of grain sterility. Regardless of fertilizer treatments, the implementation of Alternative Wetting and Drying (AWD) was the best approach to increase number of panicle per plant. The application of foliar micronutrients fertilizer did not exerted any influence on morphological parameters. Most importantly, effects of FYM were observed about changes in *Eh* (redox potential) and Zn availability. The availability of Fe and Mn were higher associated with the duration of anaerobic conditions and low *Eh.* While Cu showed its better availability at drained treatments (Wc3, Wc4) alongside FYM and FYM+NPK treatments. FYM also increased Mn availability at Wc4 condition. Fe concentration in grain increased indistinctly of fertilized treatments over Control. Although the application of FYM slightly improved the concentration of Fe, Zn, and Cu in grain, water management was the main factor controlling Zn, Cu and Mn in grain. Foliar micronutrient fertilization showed a slight increase only for Zn and Cu grain concentration.

Selection of high micronutrient crop varieties is an important tool to provide more nutrients into human diets. The result of our survey in 46 genotypes revealed that concentration of any essential nutrient widely varies among genotypes; which confirms the existence of genetic variability. Upland varieties were significantly higher in Fe, K and P in comparison to lowland varieties. Even though other elements such as Zn, Ca, Mg and Al achieved a non-depreciable increment in upland genotypes, no significance differences were observed between both genotypes. Evaluation of some red-pigmented varieties showed high Zn concentration compared with brown-white pericarp varieties. However, the result is not conclusive and further studies are recommended on red-pigmented varieties. The result reveals that upland genotypes might have higher nutrient use efficiency than lowland genotypes. The nutrients acquiring mechanisms in upland genotypes could be retained in activated even when the environmental stress has not been provoked. An anti-nutrient (phytic acid) contents was evaluated based on "phytic acid-metal molar ratios" and its inhibitory effect on bioavailability of some multivalent ions (Fe, Zn, Cu, Mn and Ca). The result showed that lowland genotypes pose higher phytic acid inhibitory potential on Fe bioavailability than upland genotypes.

Use of proper agronomic practices in combination with the plant breeding strategies is the most viable option in order to increase in micronutrients value of crops edible parts and to overcome malnutrition in vulnerable populations. The results of present studies confirm that even high-micronutrient varieties developed under soil with micronutrient deficiencies; and their biofortified potential will not be expressed to the full extent. Therefore, if the increase of micronutrient availability and improvement of soil properties are not considered in biofortification strategies, those strategies will inevitably fail.